

FAM109B siRNA (h): sc-77296

BACKGROUND

FAM109B, also known as IPIP27B (27 kDa inositol polyphosphate phosphatase interacting protein B), Sesquipedalian-2 or Ses2, is a 259 amino acid protein belonging to the sesquipedalian family and containing one PH domain, one coiled coil, and one F&H motif. FAM109B is localized to the early and recycling endosomes, the *trans*-Golgi network and macropinosomes, but is not found in late endosomes or lysosomes. FAM109B forms homodimers and heterodimers with FAM109A, and both FAM109B and FAM109A bind to the C-terminal region of the inositol polyphosphate 5-phosphatases OCRL1 and Inpp5b. Defects in the interaction of FAM109A and B with OCRL1 and Inpp5b is thought to play a role in the pathology of Lowe syndrome, characterized by ocular, renal and nervous system defects, and type 2 Dent disease, characterized by renal defects.

REFERENCES

1. Dunham, I., et al. 1999. The DNA sequence of human chromosome 22. *Nature* 402: 489-495.
2. Collins, J.E., et al. 2004. A genome annotation-driven approach to cloning the human ORFeome. *Genome Biol.* 5: R84.
3. Barbe, L., et al. 2008. Toward a confocal subcellular atlas of the human proteome. *Mol. Cell. Proteomics* 7: 499-508.
4. Swan, L.E., et al. 2010. Two closely related endocytic proteins that share a common OCRL-binding motif with APPL1. *Proc. Natl. Acad. Sci. USA* 107: 3511-3516.
5. Noakes, C.J., et al. 2011. The PH domain proteins IPIP27A and B link OCRL1 to receptor recycling in the endocytic pathway. *Mol. Biol. Cell* 22: 606-623.

CHROMOSOMAL LOCATION

Genetic locus: FAM109B (human) mapping to 22q13.2.

PRODUCT

FAM109B siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FAM109B shRNA Plasmid (h): sc-77296-SH and FAM109B shRNA (h) Lentiviral Particles: sc-77296-V as alternate gene silencing products.

For independent verification of FAM109B (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-77296A, sc-77296B and sc-77296C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

FAM109B siRNA (h) is recommended for the inhibition of FAM109B expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

FAM109B (D-2): sc-377483 is recommended as a control antibody for monitoring of FAM109B gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FAM109B gene expression knockdown using RT-PCR Primer: FAM109B (h)-PR: sc-77296-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.